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Leucotrichoic acid, a novel sesquiterpene from *Sinningia leucotricha* (Gesneriaceae)

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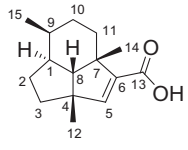
ABSTRACT

Chemical investigation of *Sinningia leucotricha* (Gesneriaceae) resulted in the isolation of leucotrichoic acid, a sesquiterpene with a novel triquinane skeleton, and the known triterpenes hederagenin and 23-hydroxyursolic acid. The chemical structures of all compounds were elucidated through spectroscopic analysis (NMR and MS). The conformational stability of the new compound was analyzed by density theory functional methods. A possible biogenesis for the new compound is proposed. Leucotrichoic acid was inactive against a panel of human cancer cell lines.

Sinningia (Gesneriaceae) is a Neotropical genus of ornamental herbs comprising 68 species, most of them native from Brazil. Several species are endangered due to intense collection and continuous decrease of their occurrence area.[1,2] *Sinningia leucotricha* (Hoehne) Moore, a plant of rare beauty, is native from Paraná State (Southern region of Brazil). Though abundant in the past, natural populations of this species are seldom found nowdays.[3] Phytochemical studies on *Sinningia* species have reported the isolation of flavonoids, steroids, phenylpropanoids, quinones, chromenes, ethylcyclohexanes, and essential oils.[4] There are no phytochemical or pharmacological studies on *S. leucotricha* yet.

This work describes the isolation and structural determination of a sesquiterpene acid (**1**) with a new carbon skeleton from the tubers of *S. leucotricha*.

Dried and powdered tubers (135 g) were extracted with ethyl acetate. The resulting extract (2.35 g) was fractionated by silica gel column and preparative thin-layer chromatography to give a mixture of hederagenin and 23-hydroxyursolic acid (5.2 mg),[5] and leucotricoic acid (**1**, 5.8 mg) as a colorless solid, $[\alpha]_D^{20}$ −33.0 (*c* 0.033, $CHCl_3$).



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Compound **1** had the molecular formula $C_{15}H_{22}O_2$, as determined from HRESIMS and NMR data, which is consistent with five degrees of unsaturation. The ^{1}H NMR spectrum (Table 1) showed signals of two tertiary methyl groups (δ_H 1.18 and 1.23) and one secondary methyl group (δ_H 0.83), several multiplets at δ_H 0.99–1.89 and, an olefinic proton (δ_H 6.71). Analysis of the ^{13}C NMR and DEPT spectra (Table 1) indicate 15 carbons, being three methyl groups (δ_C 27.4, 25.3, and 15.6), four methylene groups (δ_C 39.2, 32.3, 29.7, and 27.6), four methine carbons (δ_C 155.2, 58.0, 44.0, and 27.3), and four quaternary carbons (δ_C 170.6, 138.7, 51.8, and 47.4). These data are compatible with a tricyclic sesquiterpene acid with a double linkage. In the HMBC spectrum two methyl groups (δ_H 1.18 and 1.23, s) and the olefinic proton (H-5) showed a correlation with C-8 (δ_C 58.0), while H-5 also showed a correlation with the carbonyl group (C-13) and the methyl group at δ_H 1.18 (C-12). The overall analysis of HSQC and HMBC experiments (Fig. 1) led to structure **1**. To our knowledge this carbon skeleton, for which the name leucotrichane

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Table 1
NMR data (400 MHz, $CDCl_3$) for compound **1**

Position	1		HMBC
	δ_H (J in Hz)	δ_C	
1	1.71 (1H, m)	44.0	7, 8, 15
2a	1.30 (1H, m)	27.6	4, 8
2b	1.49 (1H, m)		4, 8
3a	1.60 (1H, m)	39.2	1, 2, 4
3b	1.72 (1H, m)		2, 4, 5, 8
4		51.8	
5	6.71 (1H, d, 0.8)	155.2	3, 4, 6, 7, 8, 12, 13
6		138.7	
7		47.4	
8	1.40 (1H, dd, 12.6, 0.8)	58.0	5, 6, 9, 12, 14
9	1.89 (1H, m)	27.3	8, 10, 15
10a	0.99 (1H, m)	29.7	11
10b	1.79 (1H, m)		7, 9, 15
11	1.52 (1H, m)	32.3	7, 8, 9, 10, 14
	1.56 (1H, m)		6, 7, 8, 10, 14
12	1.18, s	27.4	3, 4, 5, 8
13		170.6	
14	1.23, s	25.3	6, 7, 8, 11
15	0.83, d(6.8)	15.6	1, 9, 10

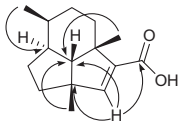

Figure 1. The most important correlations of the HMBC experiment.

is suggested, is being reported for the first time. The relative configuration of the chiral carbons (1, 4, 7, 8, and 9) in **1** was established by NOE experiments. Selective irradiation of the resonance frequency of H-8 (δ_H 1.40) caused an NOE enhancement on the signals of the methyl groups H-12, H-14, and H-15, but did not H-1. In addition, the selective irradiation of methyl groups caused an NOE enhancement on the signal of H-8. Accordingly, compound **1** has the relative configuration as represented and was named leucotrichoic acid.

Conformational analysis of **1** was performed using molecular models and density functional theory calculations (B3LYP functional as it implemented in GAUSSIAN03 suite).[6,7] According, the six-membered ring of **1** can adopt the conformation in chair or in twisted boat. In the first case, the methyl group at C-9 showed 1,3-diaxial interactions with H-8 and H-11. On the other hand,

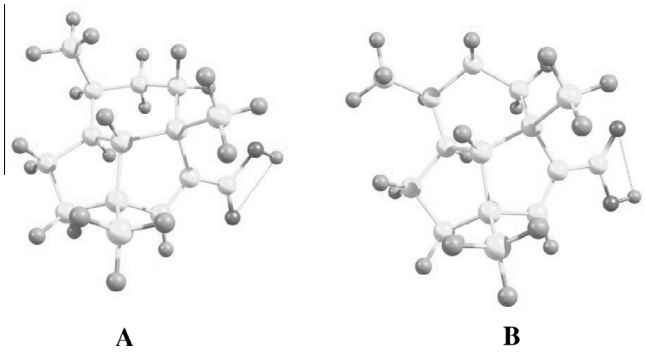

Figure 2. The main conformations of **1** (B is the less energetic).

the steric strain is alleviated in the conformation in twisted boat, which is more stable by 16 kJ mol^{-1} (Fig. 2). The theoretical optical rotation, calculated after geometrical optimization,[8] indicates the absolute configuration 1*S*, 4*R*, 7*S*, 8*S*, 9*S* for the levorotatory isomer, which was assigned for **1** according to experimental measure.

Compound **1** is structurally related to presilphiperfolanes, a small group of 5/5/6-tricyclic sesquiterpenoids found mainly in Asteraceae. The biogenesis of presilphiperfolane framework starts with farnesyl diphosphate, which after several steps leads to presilphiperfolanyl cation, a key intermediary in the biosynthesis of several compounds.[9] Compound **1** seems to derive from presilphiperfolanyl cation by a 1,2 rearrangement of C-14 from C-6 to C-7 (Scheme 1).

Compound **1** was evaluated for cytotoxic activity against nine human cancer cell lines and VERO no cancer cell line, using the method and conditions previously described,[10] but it was considered inactive because of showing values of total growth inhibition higher than 50 μg/mL against all tested cancer cell lines.

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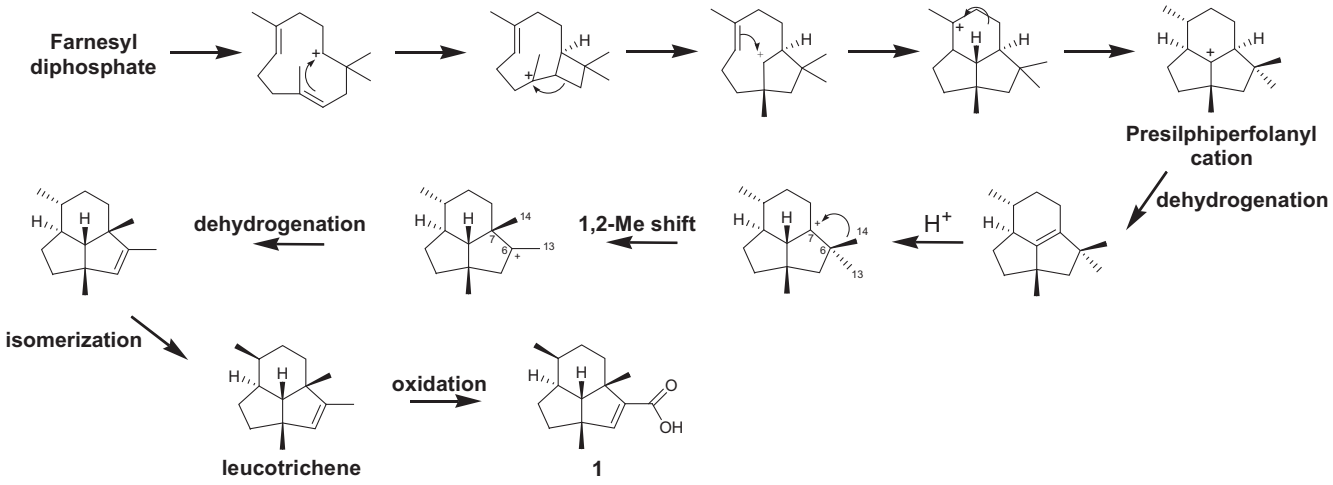
Supplementary data

Supplementary data (experimental details, 1D and 2D NMR spectra.) associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.tetlet.2013.06.108. These



Scheme 1. Possible biosynthesis of compound **1**.

data include MOL files and InChiKeys of the most important compounds described in this article.

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